Extranuclear SUMOylation in neurons

Jeremy M. Henley*, Ruth E. Carmichael and Kevin A. Wilkinson*

School of Biochemistry, Centre for Synaptic Plasticity, Biomedical Sciences Building, University of Bristol, Bristol, BS8 1TD, UK

*please address correspondence to
j.m.henley@bristol.ac.uk (Jeremy Henley)
or Kevin.Wilkinson@bristol.ac.uk (Kevin Wilkinson)

Keywords: SUMO, posttranslational modification, mitochondria, fission, synapse, AMPA receptor, kainate receptor, dynamin-related protein 1 (Drp1)

Abstract

Posttranslational modification of substrate proteins by conjugation of SUMO regulates a diverse array of cellular processes. While predominantly a nuclear protein modification, there is growing appreciation that SUMOylation of proteins outside the nucleus plays direct roles in controlling synaptic transmission, neuronal excitability and adaptive responses to cell stress. Furthermore, alterations in protein SUMOylation are observed in a wide range of neurological and neurodegenerative diseases, and several extranuclear disease-associated proteins have been shown to be directly SUMOylated. Here, focusing mainly on SUMOylation of synaptic and mitochondrial proteins, we outline recent developments and discoveries, and present our opinion as to the most exciting avenues for future research to define how SUMOylation of extranuclear proteins regulates neuronal and synaptic function.

Why is SUMOylation important?

SUMOylation is essential in nearly all eukaryotes, and it regulates the functions and fates of hundreds of proteins in a wide variety of cell pathways (for recent overview of the 'SUMOsome' see [1]). The consequences of SUMOylation are varied but the underlying principle is that it alters inter- and/or intramolecular interactions to change substrate protein localisation, stability, and/or activity. In neurons, rapid fine-tuning of protein function is essential to maintaining and modulating synaptic transmission and supporting synaptic plasticity. Importantly, a wide range of neuronal proteins, both nuclear and extranuclear, have been identified as SUMO substrates, and disrupting SUMO modification results in defects in synaptic plasticity, neuronal excitability and neuronal stress responses [2, 3]. Furthermore, a number of disease-associated proteins are SUMO substrates, and perturbations in SUMOylation have been observed in a variety of neurodegenerative disorders [2, 4]. Thus, SUMOylation has emerged as an essential regulator of neuronal function, and it is becoming increasingly apparent that SUMOylation may represent a novel therapeutic target in a wide range of diseases.

In this Opinion article, focusing mainly on synaptic and mitochondrial proteins, we present an overview of neuronal SUMOylation, outline recent developments, and discuss the major

outstanding questions relating to how SUMOylation of extranuclear proteins regulates neuronal and synaptic function.

SUMO conjugation

In mammals, there are three ~11 kDa SUMO paralogues (SUMO1-3) that covalently conjugate to lysine residues, generally within a consensus motif in target proteins [5, 6]. SUMO2 and SUMO3 are identical except for three residues, but they share only ~50% sequence identity with SUMO1 [5]. Like ubiquitination, SUMO conjugation occurs via a three-step enzymatic pathway. SUMO propeptides are processed to expose a C-terminal di-glycine motif and then 'activated' for conjugation by the E1 heterodimer of SAE1 and SAE2. Activated SUMO is passed to the E2 SUMO conjugating enzyme Ubc9 that, usually in concert with an E3 ligase, conjugates SUMO to the substrate protein [5]. In addition to being attached to substrates as monomers, SUMO proteins can also form diverse chains on substrate proteins through internal lysines, likely resulting in distinct outcomes for the modified target [2].

SUMO deconjugation

SUMOylation is a reversible modification, and SUMO can be removed from target proteins by SUMO proteases, nine of which have been identified [3] (**Box 1**). Thus, the extent and duration of substrate SUMOylation depend on a delicate balance between conjugation and SUMO protease-mediated deconjugation. Although relatively little is known about how SUMO proteases are regulated, given that there is only one E1 dimer, one E2, and a relatively small number of identified E3s for all SUMO conjugation [5, 6], it seems highly likely that the control of SUMO protease activity is a major contributor to determining how SUMOylation of individual targets, or subsets of them, is spatially and temporally regulated in response to external stimuli.

Detection of SUMOylated proteins

The identification and validation of SUMOylated target proteins is challenging due to low steady-state levels of endogenous protein modification and high levels of SUMO protease activity in cell lysates [7]. Nonetheless, advances in proteomic techniques have resulted in the identification of over 3500 substrate proteins [1], which are predominately nuclear and participate in processes such as transcription, chromatin remodelling, DNA repair and nucleocytoplasmic transport (for reviews see [8-10]).

SUMOylation of neuronal proteins outside the nucleus

As for other cell types, in neurons SUMOylation and deSUMOylation enzymes and SUMOylated target proteins are most highly enriched in the nucleus, and SUMOylation of nuclear proteins in neurons undoubtedly plays essential roles in neuronal differentiation, maturation and function [11]. However, many independent reports have confirmed that SUMOylation machinery and substrates are present and active in other neuronal compartments, including at synapses and mitochondria [2]. Indeed, numerous extranuclear proteins that are required for synaptic and mitochondrial function have been identified and functionally validated as targets of SUMOylation [2] (**Figure 1**). Nonetheless, the functional importance of SUMOylation at synapses is not universally accepted, and for instance, the existence of synaptic SUMO1-ylation in neurons has been questioned based on studies using an epitope-tagged SUMO1 knock-in mouse model [12, 13] **(Box 2**).

SUMOylation at the presynapse

Neurotransmitter release requires extremely precise spatial and temporal regulation, and manipulating presynaptic SUMOylation has profound effects on stimulation-evoked neurotransmitter release in isolated synaptosomal systems [14]. Identifying the specific presynaptic proteins that are SUMOylated in an activity-dependent and stimulus-specific manner, and defining the functional consequences for fine-tuning neurotransmitter release, is a formidable task. Nonetheless, several studies have identified individual presynaptic SUMOylation targets (**Figure 2**).

RIM1α is required for vesicle priming, Ca^{2+} channel clustering near release sites and presynaptic plasticity [15]. RIM1α was the first component of the presynaptic release machinery identified as a SUMO target and it provided important new insight into how SUMOylation can regulate synaptic function and dysfunction [16]. More specifically, RIM1α SUMOylation acts as a molecular switch; SUMOylated RIM1α clusters Ca^{2+}-channels necessary for depolarisation-evoked presynaptic Ca^{2+} signalling whereas non-SUMOylated RIM1α participates in the priming and docking of synaptic vesicles at the presynaptic active zone [16]. RIM1α also binds to **synaptotagmin-1**, a Ca^{2+} sensor in the presynaptic membrane, which is itself SUMOylated [17]. As yet, however, it is unclear if or how SUMOylation of one or both of these binding partners impacts on their interaction.

Syntaxin1A is a component of the presynaptic SNARE machinery that provides most of the mechanical force for membrane fusion and neurotransmitter release [18]. Syntaxin1A is an activity-dependent SUMO substrate and preventing its SUMOylation increases rates of vesicle endocytosis but not the rate of exocytosis, providing a flexible SUMOylation-mediated regulatory mechanism to control neurotransmitter release [19]. Furthermore, **Synapsin1a**, which anchors synaptic vesicles to the cytoskeleton and then releases them in response to depolarisation, is also a SUMO substrate. Preventing synapsin1a SUMOylation reduces the releasable vesicle pool and impairs neurotransmitter release [20].

Taken together, these data demonstrate that several key components of the release machinery are regulated by SUMOylation and we have no doubt that more will be identified and characterised. Moreover, SUMOylation is emerging as an important component of other secretory systems, particularly for insulin release from pancreatic β-cells [21].

SUMOylation of receptors and interacting proteins

Kainate receptors (KARs) are a subclass of glutamate receptors present at both the pre- and postsynaptic membranes of subsets of excitatory synapses where they regulate synaptic formation, stabilisation and function [22]. The KAR subunit GluK2 was the first reported neurotransmitter receptor protein to be SUMOylated [23] and subsequent studies have shown that its SUMOylation is regulated by prior phosphorylation by PKC, and both modifications are required for agonist-induced KAR internalization and sustained depression of KAR-mediated responses at mossy fibre-CA3 synapses [24, 25].

AMPA receptors (AMPARs) mediate most fast excitatory neurotransmission in the CNS and are critically important for nearly all aspects of brain function. In particular, regulated changes in the number and properties of AMPARs at synapses underlie long-term potentiation (LTP) and long-term depression (LTD) [26]. Although AMPARs have not been reported to be directly SUMOylated, SUMOylation plays a key role in activity-dependent AMPAR trafficking [27, 28].

For example, suppression of action potentials with TTX induces homeostatic up-scaling of AMPARs [29], and this process can be blocked by overexpression of the SENP1 catalytic domain, which reduces global SUMOylation, in cultured neurons [30]. Furthermore,

SUMOylation also plays a role in the initial insertion of AMPARs during LTP [27], since expression of the catalytic domain of SENP1, or of a dominant-negative Ubc9, blocks LTP induced chemically by application of the NMDA receptor co-agonist glycine [27]. Correspondingly, electrophysiological and *in vivo* behavioural studies have reported that acute inhibition of SUMOylation impairs LTP and hippocampal-dependent learning in mice [31] and neuron-specific knockdown of SUMO1-3 impairs episodic memory and fear conditioning, which rely on synaptic plasticity [32]. Thus, SUMOylation is required to support the AMPAR trafficking that underlies various forms of synaptic plasticity and memory formation. While these data are compelling, it is important to note that these studies relied on chronic perturbations of SUMOylation over many hours or days, so cannot conclusively distinguish whether the effects observed are due to direct modification of extranuclear or synaptic SUMO substrates, or due to changes in SUMOylation of nuclear proteins.

Nonetheless, despite the evidence supporting a role for SUMOylation in AMPAR trafficking and synaptic plasticity, the direct targets mediating these effects have largely remained elusive. An identified example is **Arc**, which plays a key role in AMPAR endocytosis in homeostatic plasticity [33]. Arc is SUMOylated [30, 34] and overexpression of wild-type Arc, but not a non-SUMOylatable Arc mutant, prevents the TTX-evoked increase in AMPAR surface expression, consistent with a role in regulating AMPAR up-scaling [30]. It has also been proposed that SUMO1-ylated Arc forms a complex with the F-actin-binding protein drebrin A, leading to a hypothesis that newly synthesized Arc is SUMOylated and targeted for regulation of the actin cytoskeleton during *in vivo* LTP [35].

SUMOylation also has a role at inhibitory synapses. **Gephyrin** is part of a scaffolding protein complex that is required for postsynaptic clustering of $GABA_A$ receptors. Gephyrin is SUMOylated and it has been reported that its deSUMOylation promotes the formation of postsynaptic clusters via cross-talk between phosphorylation and acetylation of gephyrin [36].

Group III G protein-coupled **metabotropic glutamate receptors (mGluR4, 6, 7 and 8)** are predominantly presynaptic, where they act as autoreceptors to control neurotransmitter release [37]. All of the group III mGluRs contain a SUMOylation consensus motif in their intracellular C-termini and have been reported to be substrates for SUMOylation *in vitro* [38-40]. Interestingly, in contrast to KARs, where SUMOylation is required for agonist-evoked endocytosis, it has been proposed that deSUMOylation enhances mGluR7 endocytosis, suggesting that SUMOylation stabilises the surface expression of mGluR7 in neurons [41]. However, it is noteworthy that, as for K2P1 channels (see below), this would infer that surface expressed mGluRs are stably SUMOylated, which does not fit well with the current concept of SUMOylation being a highly transient modification that acts mainly as a biological switch.

SUMOylation of ion channels

K2P1 channels provide a background potassium leak current that contributes to the resting membrane potential and reduces neuronal excitability. The K2P1 channel was the first ion channel reported to be SUMOylated, and preventing its SUMOylation unmasked K2P1 K^+ conductance, implying that constitutive SUMOylation of K2P1 silences the channel [42] (but see also [43], a study that challenges whether K2P1 is SUMOylated). As noted for mGluRs, this is unusual given that SUMOylation is generally very transient and only a small proportion of most substrates are SUMOylated at any one time [7]. Nonetheless, prompted by these reports, subsequent bioinformatics approaches revealed SUMOylation sites in

voltage-dependent potassium (K_v) channels, which also contribute to establishing resting membrane potential and to defining the duration and frequency of action potentials. SUMOylation of **K_v1.5** was shown to regulate channel inactivation [44]. In this case, preventing SUMOylation causes a selective ~15 mV hyperpolarizing shift in the voltage-dependence of steady-state inactivation with no associated effects on the voltage-dependence of activation or total current density. Thus, whereas SUMOylation of K2P1 has been reported to completely prevent K^+ conductance through the channel, SUMOylation of K_v1.5 has a more graded role. Furthermore, more recent studies have shown that SUMOylation decreases **K_v2.1** [45], **K_V7.2** [46] and **K_V7.1** [47] K^+ currents and, in contrast, SUMOylation of the voltage-dependent sodium channel **Na_V1.2** increases Na^+ currents [48]. Taken together, these reports suggest that an overarching effect of ion channel SUMOylation can be to increase neuronal excitability by suppressing K^+ channel activity as well as enhancing activity of Na^+ channels.

SUMOylation of mitochondrial proteins

Neurons are highly energy-intensive cells as a result of the demands of maintaining membrane potential and supporting the ATP-dependent neurotransmitter release and receptor recycling machinery underpinning synaptic transmission. Accordingly, neurons contain an extensive and complex mitochondrial network. Individual mitochondria undergo frequent fusion and fission events as a quality control mechanism [49]. **Drp1** is a GTPase that translocates from the cytosol to the mitochondrial outer membrane to mediate physiological fission and, under cell stress conditions, pathophysiological fragmentation and cytochrome *c* release that leads to apoptosis [50].

Current evidence suggests that SUMO1-ylation stabilizes Drp1 [51] and enhances its recruitment to mitochondria to promote fragmentation and apoptosis [52]. Conversely, SUMO2/3-ylation reduces Drp1 recruitment to the mitochondria [53], at least in part by preventing binding to its main receptor mitochondrial fission factor (Mff). Thus, SUMO2/3-ylation of Drp1 can act as an adaptive protective response in cell stress by decreasing cytochrome *c* release and apoptosis [54]. Interestingly, the extent and paralogue-specificity of Drp1 SUMOylation in neurons appears, at least in part, to be due to the activity of different SENP proteins. SENP2 knockout in mice results in neurodegeneration and mitochondrial defects, and leads to enhanced SUMO1 conjugation to Drp1 [55]. Conversely, in cultured cortical neurons, ischemic stress leads to degradation of the SUMO2/3 deconjugating enzyme SENP3, resulting in enhanced SUMOylation of Drp1 with SUMO2/3. This promotes Drp1 partitioning to the cytosol and subsequent neuronal survival [53]. Thus, the activity and levels of different SENPs can determine neuronal viability through regulating the SUMOylation status of Drp1.

While this SUMO paralogue 'switch' concept on Drp1 is attractive, it should be noted that completely preventing Drp1 SUMOylation by mutating the target lysine residues promotes mitochondrial fragmentation and cell death [53, 54]. Alternatively, it could be that that SUMO1-ylation of proteins that bind to Drp1, rather than Drp1 itself, are involved in recruitment whereas SUMO2/3-ylation of Drp1 counters mitochondrial localisation. This situation is complicated by the observation that SUMO2/3-ylation of **Fas-associated protein with Death Domain** (FADD) in stressed conditions promotes its binding to Drp1 and this, in turn, promotes Drp1 recruitment by Mff [56].

Thus, there is clearly a complex interplay of proteins and modifications that regulate Drp1-mediated mitochondrial fragmentation. It seems likely that this network of interactions

provides a mechanism to tune the actions of Drp1 to correspond to the type and severity of cell stress.

Concluding remarks

Over the last 10 years, a concerted research effort has identified crucial roles for SUMOylation in controlling neuronal excitability, synaptic plasticity and survival. While SUMOylation of nuclear targets undoubtedly plays major roles, a large number of functionally validated extranuclear SUMOylation targets have also been demonstrated to play direct roles in regulating synaptic and mitochondrial function. Moreover, aberrations in SUMOylation are associated with multiple neuropathologies [2] (Box 4). Despite this progress, there remain many unanswered questions (see Outstanding Questions).

For example, given the limited repertoire of SUMOylation enzymes, how is SUMOylation of specific substrates activity, spatially and temporally regulated to orchestrate neuronal function? More generally, the role of SUMOylation in protein network interactions is a widely applicable and largely open question. Many covalently SUMOylated proteins also possess SUMO interaction motifs (SIMs) that bind SUMO non-covalently [57] and can facilitate their binding to other SUMOylated proteins by acting as a molecular 'glue' to drive protein complex assembly. Thus, one way in which SUMOylation acts on protein networks is that, rather than proteins being individually SUMOylated, groups of proteins within a spatially defined network are SUMOylated *en masse*, a process known as 'SUMO spray' [58] (Box 3). In this scenario, the selective action of SUMO proteases could specify, collectively, the duration and location of proteins that remain SUMOylated in various cellular locations, as appears to be the case for Drp1 during ischemia [53, 54]. Moreover, given the recent questions that have arisen regarding the physiological role of SUMO1 conjugation at synapses, future studies examining the targets, regulation and paralogue specificity of SUMOylation of extranuclear substrate proteins will help clarify the breadth of roles played by SUMOylation outside the nucleus in neurons.

Finally, investigating the roles of SUMOylation in the regulation of processes that are fundamental to healthy neuronal function, and are dysregulated over the lifespan or disrupted in brain diseases, will shed new light on mechanisms underpinning healthy aging and could identify new targets for drug development to combat neurological and neurodegenerative disorders.

Glossary

AMPA receptors (AMPARs): α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptors are ionotropic glutamate receptors that mediate the vast majority of fast excitatory neurotransmission in the mammalian brain.

APP: Amyloid precursor protein (APP) is a membrane protein that can be cleaved to generate beta amyloid (Aβ). Aβ can form amyloid plaques, which are a pathological hallmark of Alzheimer's disease.

Aβ: Amyloid beta is the term encompassing a group of 36–43 amino acid peptides that are the main component of the amyloid plaques in Alzheimer's disease.

DJ-1: Protein deglycase DJ-1 (aka Parkinson disease protein 7), acts as a chaperone to inhibit α-synuclein aggregation under stressed conditions. Defects in DJ-1 are associated with early onset Parkinson's disease

Drp1: A member of the dynamin family of GTPases that plays an integral role in mitochondrial quality control, maintenance and apoptotic pathways by promoting mitochondrial fission.

FADD: Fas-associated protein with Death Domain is an adaptor protein that links specific receptors to procaspases that mediate apoptosis.

Kainate receptors (KARs): Glutamate-activated ligand-gated ion channels that play key roles in modulating neurotransmitter release at the presynapse as well as neuronal excitability and synaptic transmission at the postsynapse.

K2P1: The product of the *KCNK1* gene, K2P1 is a two-pore potassium leak channel that contributes to resting membrane potential in neurons and was the first membrane protein shown to be a SUMO substrate.

Posttranslational modification (PTM): Enzymatic attachment of chemical groups, lipids, sugars, or polypeptides to a protein after primary synthesis to modify its properties, interactions and/or functions.

RIM1α: A presynaptic active zone protein, originally identified as a Rab3A interactor, which has a role in docking and trafficking of synaptic vesicles.

SENPs: A family of proteases that remove SUMO from target proteins.

SUMO: Small ubiquitin-like modifier is a small protein that is covalently conjugated to lysines in target proteins to modify their properties.

SUMOsome: Collective noun for the proteins that are capable of being SUMOylated, generally a term used in proteomics.

Synapsin1a: A phosphoprotein that regulates the availability of synaptic vesicles for exocytosis.

Synaptotagmin-1: A presynaptic Ca^{2+} sensing protein involved in vesicle docking and release.

Syntaxin1A: A protein located at the presynaptic membrane that interacts with membrane associated SNAP-25 to form a receptor for the integral vesicle membrane protein, VAMP2, thereby forming a fundamental component of the vesicle fusion machinery.

Ubc9: The only SUMO E2 conjugating enzyme.

α-synuclein: A protein abundantly expressed in the brain that is poorly understood but that may play a role in synaptic vesicle clustering in the presynaptic terminal. Mutant forms can form insoluble fibrils that contribute to neurodegenerative conditions such as Parkinson's and Alzheimer's diseases.

Box 1

SUMO proteases

SUMOylation is a reversible covalent modification that allows for tight spatiotemporal control over substrate protein function. SUMO is deconjugated from target proteins by SUMO proteases. Nine mammalian SUMO proteases have been identified so far, but their specific targets and physiological roles remain relatively poorly characterised, and how SUMO protease activity is regulated to control substrate deSUMOylation is largely unknown (for recent reviews see [3, 59]).

The largest family of SUMO proteases are the sentrin-specific proteases (SENPs). Generally, SENP1 and SENP2 mature pre-SUMOs and deconjugate SUMO1 and SUMO2/3, SENP3 and SENP5 preferentially deconjugate SUMO2/3, and SENP6 and SENP7 can edit polySUMO2/3 chains [60].

Additionally, DeSUMOylating isopeptidases 1 and 2 (DeSI1 and DeSI2) have been identified [61]. DeSI1 seems highly specific, with only BTB-ZF identified as a substrate. The properties of DeSI2 have not been defined but, intriguingly, it exhibits a predominantly cytoplasmic rather than nuclear localisation [61]. Another recently identified but, as yet, largely uncharacterized SUMO protease is Ubiquitin-specific protease-like 1 (USPL1) [62].

What is clear, however, is that the extent and duration of substrate SUMOylation is intricately and dynamically balanced by conjugation and deconjugation. For example, regulation of the SUMO protease SENP3 directly controls the SUMO2/3-ylation status of the mitochondrial GTPase Drp1 [53, 54], supporting the concept that the stability, localisation and availability of SUMO proteases is a crucial process that determines the spatial and temporal SUMOylation of target proteins.

Thus, given that the multiple identified SUMO proteases contrast sharply with the very few core enzymes that mediate SUMO conjugation, we hypothesise that regulation of SUMO proteases is a major contributor to determining how SUMOylation of individual or subsets of targets can be controlled in response to external stimuli. Further studies into how control of SUMO protease activity orchestrates neuronal SUMOylation at various cellular locations will no doubt shed light on this question.

Box 2

Consensus and controversy: Absence of evidence or evidence of absence?

While there is a consensus that most protein SUMOylation occurs within the nucleus, multiple classes of extranuclear SUMOylated proteins have been identified, as outlined in the text, and importantly, these have been functionally validated in neurons and other cell types. The target proteins include neurotransmitter receptors and associated proteins, transporters, ion channels, key components of the presynaptic release machinery and mitochondrial proteins (for reviews see [2, 63]).

Notwithstanding these findings arising from many different labs, two related studies using a His6-HA-SUMO1 knock-in mouse model concluded that SUMO1-ylation of synaptic proteins does not occur to any significant extent [12, 13]. In the more recent paper of the two, SUMOylation of seven previously identified synaptic SUMO1 substrates was investigated – synapsin1a [20], gephyrin [36], GluK2 [23], syntaxin1a [19], RIM1α [16], mGluR7 [41], and synaptotagmin1 [17] – and no noticeable His6-HA-SUMO1-ylation of any of these proteins was detected in the His6-HA-SUMO1 knock-in mice [13]. Moreover, using subcellular fractionation and immunocytochemical staining, the authors similarly did not detect SUMO1-ylation in synaptic fractions, or colocalisation of anti-SUMO1 signal with synaptic markers.

It should be noted, however, that the mice used in these studies exhibit a 20-30% reduction in total SUMO1 conjugation [13], and – importantly, we would argue – the authors did not recapitulate the originally reported experiments in wild-type mice or rats. Indeed, it remains possible that the reduction in SUMO1 conjugation observed in these mice may be compensated for by SUMO2/3 conjugation, as occurs in SUMO1 knockout mice [64] – however, this possibility was not examined in the recent study [13]. We also note that the original studies reporting SUMOylation of synapsin1a [20], gephyrin [36], GluK2 [23], syntaxin1a [19], RIM1α [16] and mGluR7 [41] investigated the functional effects of SUMOylation. Thus, in addition to demonstrating SUMOylation *biochemically*, those studies demonstrated that SUMOylation affects the target *functionally,* something which was not addressed in the recent study questioning the functional role of SUMOylation at synapses.

As set out in more detail elsewhere [65, 66], it seems likely that an underlying reason for the lack of synaptic protein SUMO1-ylation observed in the His6-HA-SUMO1 knock-in mouse studies is that this model system does not accurately report endogenous wild-type SUMO1-ylation. The decrease in SUMO1 conjugation in the His6-HA-SUMO1 knock-in mice questions the assertion that there are no synaptic SUMO1-ylated proteins, and given these caveats, we believe these studies are not sufficiently definitive to cast doubt on the multiple lines of evidence pointing to the existence and functional relevance of synaptic SUMO1-ylation.

Box 3

SUMO spray

An intriguing concept based on quantitative mass spectrometry from experiments primarily investigating DNA repair is that SUMOylation can occur in parallel on individual proteins within a complex, or in the same immediate vicinity [58]. This has been termed “SUMO spray’ and may act to modulate multiple components of protein assemblies simultaneously. When this effect acts to hold complexes together it has been called “SUMO velcro”. This is an attractive hypothesis because it bypasses the requirement for specific SUMOylation of particular proteins in a particular compartment, which is difficult to reconcile with the fact that there is only one E1 dimer and one E2 enzyme for SUMO1- and SUMO2/3-ylation. That is, if SUMOylation pathways are available and activated, presumably all appropriate target proteins in the vicinity will be modified.

There is precedent for this idea in DNA double-strand break repair since DNA damage triggers a SUMOylation wave, leading to simultaneous multisite modifications of several repair proteins in the same pathway. This synergistic SUMOylation of several proteins stabilizes physical interactions to promote efficient DNA repair in response to specific triggers [58].

It is tempting to extend this SUMO spray concept to extranuclear compartments such as the pre- and postsynapse and mitochondria, where functional clusters or networks of proteins are likely to be subject to simultaneous SUMOylation. For example, as outlined in the main text, at the presynapse, neurotransmitter release requires highly coordinated protein-protein interactions and multiple presynaptic proteins are SUMO substrates. Moreover, SUMOylation of Drp1 at mitochondria could be accompanied by SUMOylation of other proteins in the Drp1 network that, together, regulate mitochondrial fission, particularly under stressed conditions. Indeed, it has been reported that the FADD protein is SUMOylated and it is possible that other SUMOylated proteins in this pathway will be identified in the future. Although appealing, this concept is currently difficult to assess at mitochondria or at the presynapse because there is an insufficient arsenal of tools to monitor the SUMOylation status of multiple proteins in unison. Nonetheless, we expect rapid progress in this area and that it will soon be possible to investigate how SUMOylation of groups of proteins is orchestrated to exert adaptive responses such as in synaptic plasticity and cytoprotection against severe cellular stress.

Box 4

SUMOylation and disease

SUMOylation plays key roles in regulating processes that are fundamental to healthy neuronal function. The converse is that dysregulation of protein SUMOylation is strongly implicated in neurological and neurodegenerative diseases. Furthermore, there is mounting evidence that cell stress-induced increases in protein SUMO2/3-ylation may represent an adaptive neuroprotective response (for recent reviews see [2, 67]).

An example of this is the growing consensus that SUMOylation plays an important neuroprotective role in response to ischemia. Animals that hibernate endure prolonged ischaemia and subsequent reperfusion but emerge undamaged. Intriguingly, SUMOylation is massively increased during torpor [68] and in ischaemia, and this has been proposed as a cytoprotective response [68-70]. One mechanism appears to be stress-related modulation of SENP3, which regulates the SUMOylation status and mitochondrial recruitment of Drp1, a protein that plays a key role in ischemia and reperfusion injury [3, 53], such as occurs in stroke and preeclampsia, that can cause foetal brain damage, epilepsy and autism [71].

SUMOylation of mitochondrial proteins is also directly relevant to Parkinson's disease (PD), Alzheimer's disease (AD) and other dementias in which mitochondrial dysfunction is a major factor [72, 73]. Three proteins implicated in familial PD (α-synuclein [74], DJ-1 [75] and Parkin [76]) have each been reported to be either covalently SUMOylated or associate with SUMO. These proteins regulate mitochondrial dynamics [77] and, interestingly, ablation of DJ-1 enhances SUMO-1-ylation of Drp1, leading to excessive mitochondrial fission, suggesting DJ-1 may be protective by regulating Drp1 SUMOylation [78]. Dysregulation of SUMOylation has been reported in AD and the Tg2576 transgenic AD mouse model [79], and the AD-associated proteins APP and tau have been reported to be directly SUMOylated [80-83]. Furthermore, Aβ oligomers impair activity-dependent upregulation of SUMOylation, and enhancing SUMOylation rescues Aβ-induced deficits in LTP and learning and memory tasks [79].

A characteristic factor in multiple neurodegenerative disorders is the accumulation and aggregation of disease-associated proteins in affected neurons and, in many cases, SUMOylation of the associated protein has been implicated in regulating its stability and solubility [84]. For example, α-synuclein accumulates into Lewy Bodies in PD and associated disorders, and SUMOylation of α-synuclein is strongly implicated in regulating α-synuclein solubility, although whether it promotes or reduces aggregation remains controversial [74, 85, 86]. Furthermore, PolyQ expansions in disease-associated proteins lead to the formation of aggregates in Huntington's Disease [87-89], spinobulbar muscular atrophy [90], dentatorubral-pallidoluysian atrophy [91] and a number of spinocerebellar ataxias [92-94], and several of the affected proteins have been reported to be modified by SUMO, resulting in altered solubility of the target protein.

Taken together, this large and growing body of evidence suggests that SUMOylation plays critical, but as yet ill-defined roles in the onset, progression and manifestation of many different diseases. Defining the precise mechanisms involved and generating novel therapeutic tools to modulate these systems is an exciting and highly promising area for future research.

	Protein Function	Role of SUMOylation	References
Presynaptic Proteins			
RIM1α	Presynaptic protein required for vesicle priming and calcium channel clustering near release sites	Enhances the ability of RIM1α to cluster calcium channels	[16]
Synaptotagmin-1	Presynaptic calcium sensor involved in vesicle docking and neurotransmitter release	Unknown	[17]
Syntaxin1A	Presynaptic SNARE protein that provides mechanical force for fusion of synaptic vesicles with the plasma membrane	Alters the balance of synaptic vesicle endo- and exocytosis, resulting in an increase in endocytosis	[19]
Synapsin1a	Anchors synaptic vesicles to the cytoskeleton to maintain the reserve vesicle pool	Enhances SynIa association with synaptic vesicles to promote the efficient reclustering of SynIa following neuronal stimulation	[20]
Group III mGluRs (mGluRs 4, 6, 7 and 8)	Presynaptic G-protein-coupled glutamate receptors	In the case of mGluR7, stabilizes mGluR7 surface expression.	[38-41]
Postsynaptic Proteins			
GluK2	Component of kainate-type glutamate receptors	Promotes endocytosis of GluK2-containing receptors in response to agonist	[23]
Arc	Cytoskeletal-associated protein involved in endocytosis of AMPARs	Arc SUMOylation is required for homeostatic upscaling of AMPARs. Promotes association of Arc with the actin-binding protein Drebrin A	[30, 35]
Gephyrin	Postsynaptic scaffold at inhibitory synapses that plays a critical role in scaffolding $GABA_A$ receptors	Antagonises the formation of postsynaptic gephyrin clusters	[36]
Ion Channels			

K2P1	Potassium leak channel	SUMOylation blocks ion flow through the channel	[42]
$K_v1.5$	Voltage-gated potassium channel	Affects the voltage-dependence of steady-state inactivation	[44]
$K_v2.1$	Voltage-gated potassium channel	Increases the half-maximal activation voltage, reducing channel activity	[45]
$K_V7.1$	Voltage-gated potassium channel	Increases the half-maximal activation voltage, reducing channel activity	[47]
$K_V7.2$	Voltage-gated potassium channel	Reduces the $K_V7.2$-dependent hyperpolarizing M-current	[46]
$Na_V1.2$	Voltage-gated sodium channel	Affects voltage-dependence of channel activation, resulting in increased sodium currents	[48]
Mitochondrial Proteins			
Drp1	GTPase that mediates mitochondrial fission	Controls Drp1 partitioning to mitochondria; SUMO1-ylation promotes mitochondrial recruitment, SUMO2/3-ylation favours cytosolic localization	[51-53]
FADD	Adaptor protein involved in the induction of apoptosis	Enhances binding to Drp1, promoting recruitment of Drp1 to mitochondria during cell stress	[56]

Table 1: Table summarising the SUMO substrates discussed in the text.

Figure Legends

Figure 1. Subcellular localisation of SUMO machinery and SUMO targets in neurons. Substrate proteins for SUMOylation and components of the SUMOylation machinery can be found throughout neurons. The schematic highlights where components of the SUMO machinery have been shown to be localised (left hand dotted box), and the diverse cellular locations of known SUMO substrates (right hand dotted box). While the majority of known SUMO substrates are nuclear, pre- and postsynaptic proteins are also SUMOylated (see also Figure 2). At the presynapse, SUMOylation alters the function of various proteins involved in neurotransmitter release, to control the efficacy of synaptic transmission. At the postsynapse, SUMOylation regulates the activity of receptors and scaffolding proteins. Moreover, SUMOylation of several classes of ion channels localised at the plasma membrane alters their activity and tunes neuronal excitability. Finally, SUMOylation of the mitochondrial proteins Drp1 and FADD plays roles in mitochondrial dynamics and in neuronal responses to stress. For more details see also Table 1. Figure generated from shapes available from Servier Medical Art (https://smart.servier.com/) under a Creative Commons Attribution 3.0 Unported License.

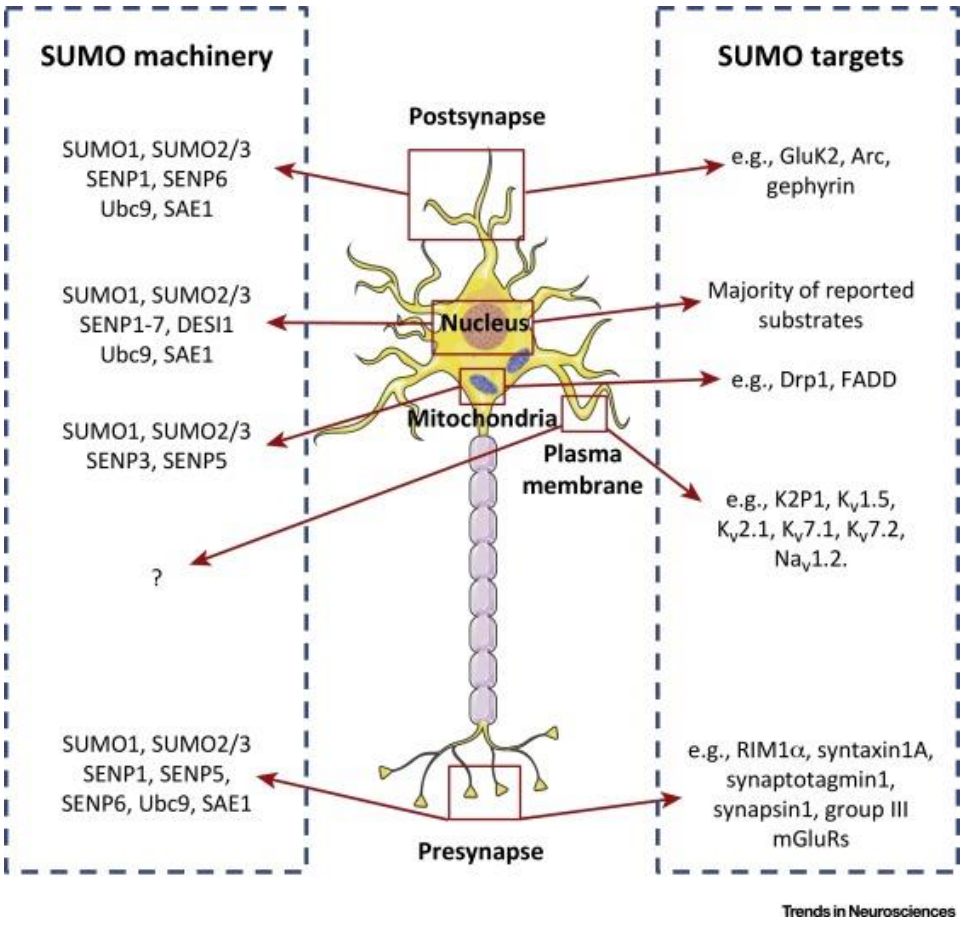
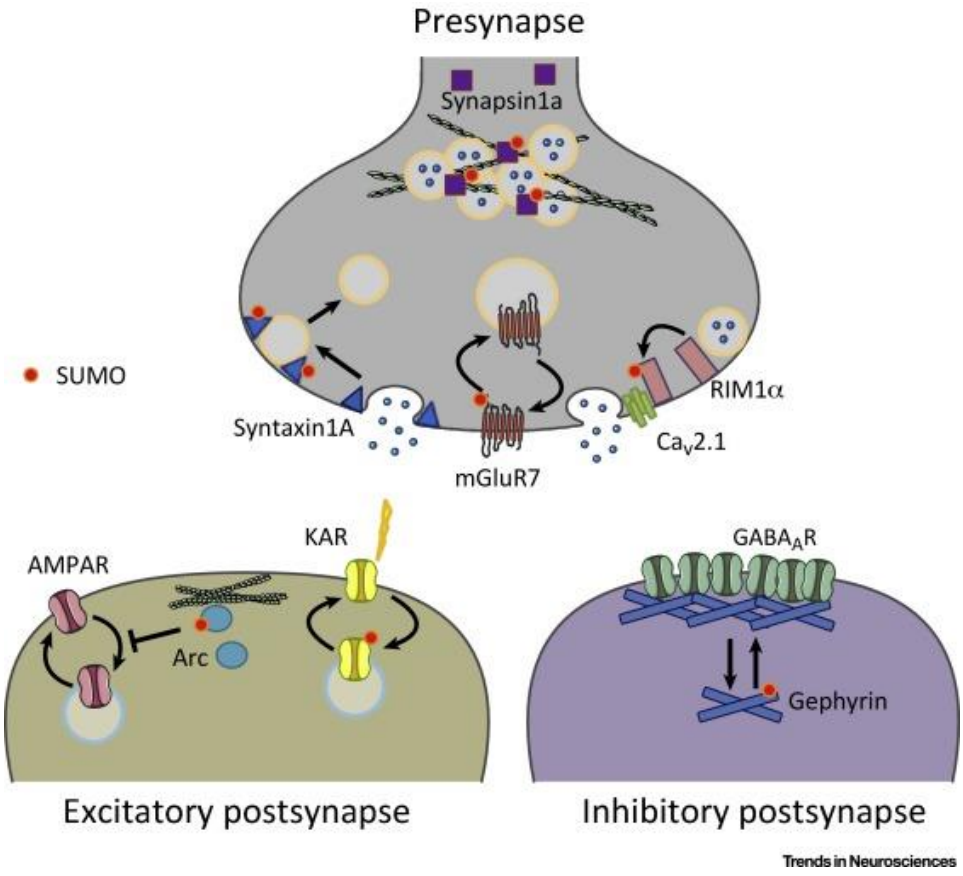


Figure 2. Functional consequences of synaptic SUMOylation

SUMOylation and deSUMOylation of target proteins at synapses has profound effects on synaptic function. Known SUMO substrates localised to the presynapse (top), as well as to excitatory or inhibitory postsynapses (bottom left/right, respectively) are shown. SUMO-modified species are represented with a red SUMO moiety attached. In the schematic axon terminal and presynapse (top), known SUMO substrates are highlighted. SUMOylation of RIM1α alters its function by facilitating calcium channel clustering to promote neurotransmitter release. SUMOylation of Synapsin1a is required to maintain the cytoskeleton-associated releasable pool of synaptic vesicles by enhancing the interaction between Synapsin1a and the synaptic vesicles, while Syntaxin1a SUMOylation increases the rate of vesicle endocytosis at the presynaptic membrane. Furthermore, the presynaptic glutamate receptor mGluR7 is stabilised at the cell surface by SUMO modification.

At excitatory postsynapses (bottom left), SUMOylation of Arc enhances its recruitment to the cytoskeleton via the actin-binding protein drebrin A and reduces its ability to promote AMPAR endocytosis. The kainate receptor subunit GluK2 is SUMOylated upon agonist binding, driving its internalisation to depress KAR-mediated synaptic responses. At inhibitory postsynapses (bottom right), gephyrin deSUMOylation favours the formation of postsynaptic $GABA_AR$ clusters. Figure generated from shapes available from Servier Medical Art (https://smart.servier.com/) under a Creative Commons Attribution 3.0 Unported License.



.

Acknowledgements

We are grateful to the MRC, BBSRC, Parkinson's UK, British Heart Foundation, Alzheimer's Research UK and BRACE for funding. Shapes used in Figures 1 and 2 were adapted from Servier Medical Art (https://smart.servier.com/) licensed under a Creative Commons Attribution 3.0 Unported License. The authors confirm they have no conflicts of interest related to this work.

References

1. Hendriks, I.A. and Vertegaal, A.C. (2016) A comprehensive compilation of SUMO proteomics. Nat Rev Mol Cell Biol.
2. Henley, J.M. et al. (2014) Neuronal SUMOylation: mechanisms, physiology, and roles in neuronal dysfunction. Physiol Rev 94 (4), 1249-85.
3. Guo, C. and Henley, J.M. (2014) Wrestling with stress: roles of protein SUMOylation and deSUMOylation in cell stress response. IUBMB Life 66 (2), 71-7.
4. Anderson, D.B. et al. (2017) Sumoylation: Implications for Neurodegenerative Diseases. Adv Exp Med Biol 963, 261-281.
5. Wilkinson, K.A. and Henley, J.M. (2010) Mechanisms, regulation and consequences of protein SUMOylation. Biochem J 428 (2), 133-45.
6. Gareau, J.R. and Lima, C.D. (2011) The SUMO pathway: emerging mechanisms that shape specificity, conjugation and recognition. Nat Rev Mol Cell Biol 11 (12), 861-71.
7. Hay, R.T. (2005) SUMO: a history of modification. Mol Cell 18 (1), 1-12.
8. Bergink, S. and Jentsch, S. (2009) Principles of ubiquitin and SUMO modifications in DNA repair. Nature 458 (7237), 461-7.
9. Heun, P. (2007) SUMOrganization of the nucleus. Curr Opin Cell Biol.
10. Flotho, A. and Melchior, F. (2013) Sumoylation: a regulatory protein modification in health and disease. Annu Rev Biochem 82, 357-85.
11. Yamada, T. et al. (2013) Sumoylated MEF2A coordinately eliminates orphan presynaptic sites and promotes maturation of presynaptic boutons. J Neurosci 33 (11), 4726-40.
12. Tirard, M. et al. (2012) In vivo localization and identification of SUMOylated proteins in the brain of His6-HA-SUMO1 knock-in mice. Proc Natl Acad Sci U S A 109 (51), 21122-7.
13. Daniel, J.A. et al. (2017) Analysis of SUMO1-conjugation at Synapses. Elife 6.
14. Feligioni, M. et al. (2009) Protein SUMOylation modulates calcium influx and glutamate release from presynaptic terminals. Eur J Neurosci 29 (7), 1348-56.
15. Muller, M. et al. (2012) RIM controls homeostatic plasticity through modulation of the readily-releasable vesicle pool. J Neurosci 32 (47), 16574-85.
16. Girach, F. et al. (2013) RIM1alpha SUMOylation Is Required for Fast Synaptic Vesicle Exocytosis. Cell Rep 5 (5), 1294-301.
17. Matsuzaki, S. et al. (2015) SUMO1 Affects Synaptic Function, Spine Density and Memory. Sci Rep 5, 10730.
18. Risselada, H.J. and Grubmuller, H. (2012) How SNARE molecules mediate membrane fusion: recent insights from molecular simulations. Curr Opin Struct Biol 22 (2), 187-96.
19. Craig, T.J. et al. (2015) SUMOylation of Syntaxin1A regulates presynaptic endocytosis. Sci Rep 5, 17669.
20. Tang, L.T. et al. (2015) SUMOylation of synapsin Ia maintains synaptic vesicle availability and is reduced in an autism mutation. Nat Commun 6, 7728.
21. Ferdaoussi, M. et al. (2017) SUMOylation and calcium control syntaxin-1A and secretagogin sequestration by tomosyn to regulate insulin exocytosis in human ss cells. Sci Rep 7 (1), 248.
22. Palacios-Filardo, J. et al. (2016) Synaptic Targeting of Kainate Receptors. Cereb Cortex 26 (4), 1464-72.
23. Martin, S. et al. (2007) SUMOylation regulates kainate-receptor-mediated synaptic transmission. Nature 447 (7142), 321-5.
24. Konopacki, F.A. et al. (2011) Agonist-induced PKC phosphorylation regulates GluK2 SUMOylation and kainate receptor endocytosis. Proc Natl Acad Sci U S A 108 (49), 19772-7.
25. Chamberlain, S.E. et al. (2012) SUMOylation and phosphorylation of GluK2 regulate kainate receptor trafficking and synaptic plasticity. Nat Neurosci 15 (6), 845-52.
26. Henley, J.M. and Wilkinson, K.A. (2016) Synaptic AMPA receptor composition in development, plasticity and disease. Nat Rev Neurosci 17 (6), 337-50.

27. Jaafari, N. et al. (2013) SUMOylation is required for glycine-induced increases in AMPA receptor surface expression (ChemLTP) in hippocampal neurons. PLoS One 8 (1), e52345.
28. Craig, T.J. et al. (2012) Homeostatic Synaptic Scaling Is Regulated by Protein SUMOylation. J Biol Chem 287 (27), 22781-8.
29. Turrigiano, G. (2012) Homeostatic synaptic plasticity: local and global mechanisms for stabilizing neuronal function. Cold Spring Harb Perspect Biol 4 (1), a005736.
30. Craig, T.J. et al. (2012) Homeostatic Synaptic Scaling Is Regulated by Protein SUMOylation. J Biol Chem 287 (27), 22781-8.
31. Lee, D. et al. (2012) Hippocampal place fields emerge upon single-cell manipulation of excitability during behavior. Science 337 (6096), 849-53.
32. Wang, L. et al. (2014) Neuron-specific Sumo1-3 knockdown in mice impairs episodic and fear memories. J Psychiatry Neurosci 39 (4), 259-66.
33. Chowdhury, S. et al. (2006) Arc/Arg3.1 interacts with the endocytic machinery to regulate AMPA receptor trafficking. Neuron 52 (3), 445-59.
34. Bramham, C.R. et al. (2008) The immediate early gene arc/arg3.1: regulation, mechanisms, and function. J Neurosci 28 (46), 11760-7.
35. Nair, R.R. et al. (2017) Dynamic Arc SUMOylation and Selective Interaction with F-Actin-Binding Protein Drebrin A in LTP Consolidation In Vivo. Front Synaptic Neurosci 9, 8.
36. Ghosh, H. et al. (2016) Several posttranslational modifications act in concert to regulate gephyrin scaffolding and GABAergic transmission. Nat Commun 7, 13365.
37. Enz, R. (2007) The trick of the tail: protein-protein interactions of metabotropic glutamate receptors. Bioessays 29 (1), 60-73.
38. Wilkinson, K.A. et al. (2008) Analysis of SUMO-1 modification of neuronal proteins containing consensus SUMOylation motifs. Neurosci Lett 436 (2), 239-44.
39. Tang, Z. et al. (2005) Pias1 interaction and sumoylation of metabotropic glutamate receptor 8. J Biol Chem 280 (46), 38153-9.
40. Dutting, E. et al. (2011) SUMO E3 ligases are expressed in the retina and regulate SUMOylation of the metabotropic glutamate receptor 8b. Biochem J 435 (2), 365-71.
41. Choi, J.H. et al. (2016) Regulation of mGluR7 trafficking by SUMOylation in neurons. Neuropharmacology 102, 229-35.
42. Rajan, S. et al. (2005) Sumoylation silences the plasma membrane leak K+ channel K2P1. Cell 121 (1), 37-47.
43. Feliciangeli, S. et al. (2007) Does sumoylation control K2P1/TWIK1 background K+ channels? Cell 130 (3), 563-9.
44. Benson, M.D. et al. (2007) SUMO modification regulates inactivation of the voltage-gated potassium channel Kv1.5. Proc Natl Acad Sci U S A 104 (6), 1805-10.
45. Plant, L.D. et al. (2011) SUMO modification of cell surface Kv2.1 potassium channels regulates the activity of rat hippocampal neurons. J Gen Physiol 137 (5), 441-54.
46. Qi, Y. et al. (2014) Hyper-SUMOylation of the Kv7 potassium channel diminishes the M-current leading to seizures and sudden death. Neuron 83 (5), 1159-71.
47. Xiong, D. et al. (2017) SUMOylation determines the voltage required to activate cardiac IKs channels. Proc Natl Acad Sci U S A 114 (32), E6686-E6694.
48. Plant, L.D. et al. (2016) SUMOylation of NaV1.2 channels mediates the early response to acute hypoxia in central neurons. Elife 5.
49. Chan, D.C. (2012) Fusion and fission: interlinked processes critical for mitochondrial health. Annu Rev Genet 46, 265-87.
50. Detmer, S.A. and Chan, D.C. (2007) Functions and dysfunctions of mitochondrial dynamics. Nat Rev Mol Cell Biol 8 (11), 870-9.
51. Harder, Z. et al. (2004) Sumo1 conjugates mitochondrial substrates and participates in mitochondrial fission. Curr Biol 14 (4), 340-5.
52. Prudent, J. et al. (2015) MAPL SUMOylation of Drp1 Stabilizes an ER/Mitochondrial Platform Required for Cell Death. Mol Cell 59 (6), 941-55.
53. Guo, C. et al. (2013) SENP3-mediated deSUMOylation of dynamin-related protein 1 promotes cell death following ischaemia. EMBO J 32 (11), 1514-28.

54. Guo, C. et al. (2017) SENP3-mediated deSUMOylation of Drp1 facilitates interaction with Mff to promote cell death. Sci Rep 7, 43811.
55. Fu, J. et al. (2014) Disruption of SUMO-specific protease 2 induces mitochondria mediated neurodegeneration. PLoS Genet 10 (10), e1004579.
56. Choi, S.G. et al. (2016) SUMO-Modified FADD Recruits Cytosolic Drp1 and Caspase-10 to Mitochondria for Regulated Necrosis. Mol Cell Biol.
57. Kerscher, O. (2007) SUMO junction-what's your function? New insights through SUMO-interacting motifs. EMBO Rep 8 (6), 550-5.
58. Psakhye, I. and Jentsch, S. (2012) Protein group modification and synergy in the SUMO pathway as exemplified in DNA repair. Cell 151 (4), 807-20.
59. Hickey, C.M. et al. (2012) Function and regulation of SUMO proteases. Nat Rev Mol Cell Biol 13 (12), 755-66.
60. Bekes, M. et al. (2011) The dynamics and mechanism of SUMO chain deconjugation by SUMO-specific proteases. J Biol Chem 286 (12), 10238-47.
61. Shin, E.J. et al. (2012) DeSUMOylating isopeptidase: a second class of SUMO protease. EMBO Rep 13 (4), 339-46.
62. Schulz, S. et al. (2012) Ubiquitin-specific protease-like 1 (USPL1) is a SUMO isopeptidase with essential, non-catalytic functions. EMBO Rep 13 (10), 930-8.
63. Schorova, L. and Martin, S. (2016) Sumoylation in Synaptic Function and Dysfunction. Front Synaptic Neurosci 8, 9.
64. Evdokimov, E. et al. (2008) Loss of SUMO1 in mice affects RanGAP1 localization and formation of PML nuclear bodies, but is not lethal as it can be compensated by SUMO2 or SUMO3. J Cell Sci 121 (Pt 24), 4106-13.
65. Wilkinson, K.A. et al., Is the His6-HA-SUMO1 knock-in mouse a valid model system to study protein SUMOylation? Comment on PMID 28598330: Analysis of SUMO1-conjugation at synapses., PubMed Commons, Bethesda (MD): National Library of Medicine, 2017, p. https://www.ncbi.nlm.nih.gov/pubmed/28598330#comments.
66. Wilkinson, K.A. et al. (2017) Commentary: Analysis of SUMO1-conjugation at synapses. Frontiers in Cellular Neuroscience 11 (345).
67. Liu, F.Y. et al. (2017) SUMOylation in Neurological Diseases. Curr Mol Med.
68. Lee, Y.J. et al. (2007) Protein SUMOylation is massively increased in hibernation torpor and is critical for the cytoprotection provided by ischemic preconditioning and hypothermia in SHSY5Y cells. J Cereb Blood Flow Metab 27 (5), 950-62.
69. Datwyler, A.L. et al. (2011) SUMO2/3 conjugation is an endogenous neuroprotective mechanism. J Cereb Blood Flow Metab 31 (11), 2152-9.
70. Cimarosti, H. et al. (2012) Enhanced SUMOylation and SENP-1 protein levels following oxygen and glucose deprivation in neurones. J Cereb Blood Flow Metab 32 (1), 17-22.
71. Baczyk, D. et al. (2013) Emerging role of SUMOylation in placental pathology. Placenta 34 (7), 606-12.
72. Lim, K.L. et al. (2006) Parkin-mediated lysine 63-linked polyubiquitination: a link to protein inclusions formation in Parkinson's and other conformational diseases? Neurobiol Aging 27 (4), 524-9.
73. Sheng, Z.H. and Cai, Q. (2012) Mitochondrial transport in neurons: impact on synaptic homeostasis and neurodegeneration. Nat Rev Neurosci 13 (2), 77-93.
74. Krumova, P. et al. (2011) Sumoylation inhibits {alpha}-synuclein aggregation and toxicity. J Cell Biol 194 (1), 49-60.
75. Shinbo, Y. et al. (2006) Proper SUMO-1 conjugation is essential to DJ-1 to exert its full activities. Cell Death Differ 13 (1), 96-108.
76. Um, J.W. and Chung, K.C. (2006) Functional modulation of parkin through physical interaction with SUMO-1. J Neurosci Res 84 (7), 1543-54.
77. Wang, X. et al. (2012) Parkinson's disease-associated DJ-1 mutations impair mitochondrial dynamics and cause mitochondrial dysfunction. J Neurochem 121 (5), 830-9.
78. Shimizu, Y. et al. (2016) DJ-1 protects the heart against ischemia-reperfusion injury by regulating mitochondrial fission. J Mol Cell Cardiol 97, 56-66.

79. Lee, L. et al. (2014) Regulation of synaptic plasticity and cognition by SUMO in normal physiology and Alzheimer's disease. Sci Rep 4, 7190.
80. Luo, H.B. et al. (2014) SUMOylation at K340 inhibits tau degradation through deregulating its phosphorylation and ubiquitination. Proc Natl Acad Sci U S A 111 (46), 16586-91.
81. Takahashi, K. et al. (2008) SUMO-1 immunoreactivity co-localizes with phospho-Tau in APP transgenic mice but not in mutant Tau transgenic mice. Neurosci Lett 441 (1), 90-3.
82. Dorval, V. et al. (2007) Modulation of Abeta generation by small ubiquitin-like modifiers does not require conjugation to target proteins. Biochem J 404 (2), 309-16.
83. Dorval, V. and Fraser, P.E. (2006) Small ubiquitin-like modifier (SUMO) modification of natively unfolded proteins tau and alpha-synuclein. J Biol Chem 281 (15), 9919-24.
84. Liebelt, F. and Vertegaal, A.C. (2016) Ubiquitin-Dependent and Independent Roles of SUMO in Proteostasis. Am J Physiol Cell Physiol, ajpcell 00091 2016.
85. Abeywardana, T. and Pratt, M.R. (2015) Extent of inhibition of alpha-synuclein aggregation in vitro by SUMOylation is conjugation site- and SUMO isoform-selective. Biochemistry 54 (4), 959-61.
86. Rott, R. et al. (2017) SUMOylation and ubiquitination reciprocally regulate alpha-synuclein degradation and pathological aggregation. Proc Natl Acad Sci U S A 114 (50), 13176-13181.
87. Steffan, J.S. et al. (2004) SUMO modification of Huntingtin and Huntington's disease pathology. Science 304 (5667), 100-4.
88. Subramaniam, S. et al. (2009) Rhes, a striatal specific protein, mediates mutant-huntingtin cytotoxicity. Science 324 (5932), 1327-30.
89. O'Rourke, J.G. et al. (2013) SUMO-2 and PIAS1 modulate insoluble mutant huntingtin protein accumulation. Cell Rep 4 (2), 362-75.
90. Mukherjee, S. et al. (2009) Small ubiquitin-like modifier (SUMO) modification of the androgen receptor attenuates polyglutamine-mediated aggregation. J Biol Chem 284 (32), 21296-306.
91. Terashima, T. et al. (2002) SUMO-1 co-localized with mutant atrophin-1 with expanded polyglutamines accelerates intranuclear aggregation and cell death. Neuroreport 13 (17), 2359-64.
92. Guo, L. et al. (2014) A cellular system that degrades misfolded proteins and protects against neurodegeneration. Mol Cell 55 (1), 15-30.
93. Zhou, Y.F. et al. (2013) SUMO-1 modification on K166 of polyQ-expanded ataxin-3 strengthens its stability and increases its cytotoxicity. PLoS One 8 (1), e54214.
94. Janer, A. et al. (2010) SUMOylation attenuates the aggregation propensity and cellular toxicity of the polyglutamine expanded ataxin-7. Hum Mol Genet 19 (1), 181-95.